ministration of lipase[2] or of a detergent (Tween 80)[3] with the fat meal reduced the hyperchylomicronemia of older persons to practically the level of the younger age group. The chylomicron count in young persons following a fat meal was not influenced materially by the administration of lipase with the fat meal.

We have found earlier (*11*) that pancreatic lipase secretion and blood lipase levels are significantly lower in older persons than in young ones. The effect of lipase or of a detergent in reducing the hyperchylomicronemia in old subjects to levels of young subjects seems to support the assumption that the mechanisms of fat digestion or of fat absorption, probably both, change with aging. It is not probable that hyperchylomicronemia is due to delayed disposition of circulating fat, because intravenous injection of equal volumes of hyperlipemic plasma into young and old subjects yielded similar chylomicron curves (*1*). It is premature to speculate whether administration of lipase or of detergent to normal persons may prohibit the development of atherosclerosis, or whether the progression of the disease can be interrupted by the administration of lipase or of detergent. Animal experiments may answer this question.

In persons over 50 years of age ingestion of a small amount of oleomargarine was followed by a practically 24 hr increase in the chylomicron count in the serum. In younger persons the chylomicron curve returned to fasting levels within 5 hr. Since all people eat some fat at least once a day, increased numbers of fat particles circulate in the blood of older persons practically permamently. If it is true that particulate fat, circulating in the blood, leads to atherosclerosis, the condition leading to that degenerative disease has been found.

Administration of lipase or of detergent with the fat meal reduced the chylomicron counts and the duration of increased counts of old subjects to levels of young subjects.

Work on animals will show whether atherosclerosis can be influenced by drugs affecting digestion and absorption of fat.

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[2] Kindly supplied by Dr. D. Klein, Wilson Company, in the form of concentrated pancreatin.

[3] Kindly supplied by Dr. G. R. Hazel, Abbott Laboratories.

A Vibrating Tissue Slicer[1]

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Some recent studies (*5*) of the metabolism of brain cortex slices required six or eight parallel experiments on the same brain. Neither the freehand method (*4*), nor the microtome method (*3*) yields sufficient material from brains of guinea pigs or rabbits. We have found that a vibrating blade with freehand manipulation can provide 500 mg of cortex slices from a full-grown guinea pig brain. The source of vibration is a mechanical wood carving tool (Burgess Vibro-Tool), which develops a push-pull vibration at a frequency of 120 per sec. There is only slight lateral vibration. In the chuck, we fastened a split metal adapter tightened by a wing nut, which can hold 2-in. pieces of Stadie blades or single-edge razor blades.

TABLE 1

	Hand cut		Cutter	
	Wet weight in mg	Qo_2	Wet weight in mg	Qo_2
Mouse liver	122	5.77	78	6.08
	94	6.28	82	5.58
	112	5.79	92	6.06
Average		5.95		5.92
Guinea pig brain cortex	52	9.18	57	10.20
	72	9.72	71	9.29
	53	10.10	63	9.56
Average		9.67		9.68

The platform upon which the cerebral hemisphere is placed is made by filling a 50-mm crystallizing basin with ice and water to overflowing, sliding the bottom of a larger basin over it, and inverting the basins. The ice floats to the top, causing the platform to be chilled. The cerebral hemisphere, with meninges and blood vessels removed, is placed on a small square of moistened filter paper on this platform.

The cutter, with the edge facing the operator, is grasped firmly in both hands, one arm resting on the table edge for added stability. The vibrating blade is drawn slowly through the tissue, toward the operator, with a wrist motion. The slice lies flat on the blade, and the plane of the cut can be varied for thickness of slice and for contour of the tissue without the necessity of guiding and sawing simultaneously, which is inherent in the freehand method. The slice can be picked off the blade with a fine forceps or can be removed by dipping the vibrating blade into a small beaker of chilled Ringer's solution. The blade must be moistened slightly if the

[1] This work has been supported in part by the Office of Naval Research and the International Minerals and Chemical Corporation.

slicing is to be done in the open, but need not be moistened in the humid box of Sperry (*2*) or the moist cold box of Fuhrman and Field (*1*).

There is no apparent change in the tissues induced by the low frequency vibration of the blade, as measured by the 1-hr oxygen uptake of comparative portions of the same organ sliced by hand (*4*) and by the vibrating cutter, as shown in Table 1. The medium used was Kreb's Phosphate Ringer's, pH 7.4, at 37° C, with air atmosphere. The brain medium also contained .011 molar glucose.

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The Chemical Nature of a Factor in Hog Stomach Extracts that Reduces the Creatinuria of Muscular Dystrophy[1]

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A labile factor, assayed by its effect on lowering the creatinuria of a patient with progressive muscular dystrophy, has been found to concentrate in the fat fraction (*7*) of hog gastric mucin[2] and of hog stomach linings[3]. But attempts at further fractionation of this concentrate by countercurrent distribution between absolute methanol and isooctane led to a rapid loss of biological activity. The crude fat fraction contained up to 1%[4] of a substance which reduced ferric chloride in a modified Emmerie and Engel (*1*) assay. This reducing property proved to be a measure of biological activity, since loss in biological activity was associated with a decrease in amount of reducing substance. None of the four naturally occurring tocopherols, which also reduce ferric chloride in the assay of Emmerie and Engel, could have accounted for the biological activity, since they were without effect on the creatinuria of this patient at doses 25 times the active dose of reducing substance from stomach extracts. Alpha (*6*), beta, and gamma tocopherols (*4*), however, do abolish the creatinuria of the muscular dystrophy of experimental vitamin E deficiency.

The fat fraction of hog gastric mucin which lowered creatinuria at a dosage of 100 mg was allowed to lose activity by standing at room temperature for 3 months. The amount of reducing substance fell from 0.77% to 0.07% and biological activity was completely lost. This suggested a destruction by autoxidation which leads, in the case of tocopherols in the presence of fats, to the formation of tocopheryl-*p*-quinones (*6*). The tocopheryl-*p*-quinones, in turn, can be reduced to the corresponding tocopheryl-*p*-hydroquinones and cyclized in the presence of mineral acid to regenerate the original tocopherols (*5*). The following experiments, which were designed to regenerate any tocopherols in the inactivated fat fraction that had undergone autoxidation and tocopheryl-*p*-quinone formation, led to the finding that an intermediate in the reaction, a simple reduction product, possessed chemical and biological properties similar to the factor in the fat fraction of hog gastric mucin. A 100-mg portion of inactivated fat fraction was refluxed for 2 hr in an isooctane-ethanol mixture containing 5.0 g stannous chloride and 5.0 ml concentrated HCl. After addition of water and recovery of the isooctane layer, the amount of reducing substance was found to have increased to 1.08% and remained stable at this value. A 225-mg dose of this material containing 2.43 mg of reducing substance had no effect on creatinuria. A control experiment with pure alpha tocopheryl-*p*-quinone under the same conditions showed complete conversion to alpha tocopherol ($E\,^{1\%}_{1\,cm}$ [298 mμ] in isooctane = 70; $E\,^{1\%}_{1\,cm}$ [520 mμ] in the Emmerie and Engel assay = 370). Another 100-mg portion of inactivated fat fraction of gastric mucin was refluxed for 30 min in the same solvent with 0.7 g stannous chloride and 0.5 ml concentrated HCl, and the isooctane layer was recovered as described. The reducing substance had increased to 0.73% but fell to 0.30% on the second day, 0.27% on the third day, and to 0.21% by the end of a week. A 225-mg dose of this material, containing 1.03 mg of reducing substance, was biologically active. The control with pure alpha tocopheryl-*p*-quinone under these conditions showed the formation of alpha tocopheryl-*p*-hydroquinone by the appearance of an absorption maximum at 290 mμ which disappeared in the course of 18 hr, as the spectrum of alpha tocopheryl-*p*-quinone reappeared with a double maximum between 260 and 270 mμ. This rapid autoxidation is characteristic of alpha tocopheryl-*p*-hydroquinone, first described by John (*5*). The creatinuria-lowering activity of the fraction of gastric mucin treated in this way was fully duplicated by 1.0 mg of pure synthetic alpha tocopheryl-*p*-hydroquinone.

Alpha tocopheryl-*p*-hydroquinone was prepared as needed from the more stable alpha tocopheryl-*p*-quinone by catalytic hydrogenation in ethanol or propylene glycol with palladium on calcium carbonate. The $E\,^{1\%}_{1\,cm}$ (290 mμ) in isooctane was 88 immediately after hydrogenation, a value somewhat higher than that reported by John (*5*). The $E\,^{1\%}_{1\,cm}$ (520 mμ) in the Emmerie and Engel assay was

[1] This work has been aided by the Armour Fund for Research in Muscular Disease and by a grant from the Nutrition Foundation.

[2] Frederick Stearns and Company. Prepared by alcoholic precipitation of an acid digestion of hog stomach linings.

[3] A low temperature concentrate of an acetone extract of hog stomach linings was kindly furnished by the Armour Research Laboratories.

[4] Calculated from a standard curve for alpha tocopherol.